HHS Public Access

Author manuscript

Sci Total Environ. Author manuscript; available in PMC 2018 January 01.

Published in final edited form as:

Sci Total Environ. 2017 January 01; 575: 400–405. doi:10.1016/j.scitotenv.2016.10.028.

Serum polybrominated diphenyl ether (PBDE) concentrations in relation to biomarkers of oxidative stress and inflammation: The National Health and Nutrition Examination Survey 2003–2004

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Abstract

Exposure to Polybrominated diphenyl ethers (PBDEs) has been associated with various adverse health outcomes related to liver, neural and endocrine systems. Some of these may be the result of PBDE-induced oxidative stress or inflammation, but these associations have been explored minimally in humans. In the present study we examined the relationship between PBDE concentrations and biomarkers of oxidative stress and inflammation measured in blood samples among a representative US sample from the National Health and Nutrition Examination Survey. Oxidative stress biomarkers showed no significant associations with PBDEs in adjusted regression models. For inflammation biomarkers, we observed small but statistically significant positive associations between BDE-153 and alkaline phosphatase (percent change with an interquartile range [IQR] increase in BDE-153=0.82, 95% confidence interval [CI] =0.01, 1.65) and absolute neutrophil count (percent change with IQR increase in BDE-153= 0.53%, 95% CI=0.03, 1.04). Associations with other PBDE congeners and inflammation markers were generally positive but did not reach statistical significance. These results are consistent with human research of oxidative stress and inflammation in response to PBDE congeners and mixtures, and support previous reports of inflammation in response to PBDE treatment in animal and *in vitro* studies. More detailed toxicological and epidemiologic research in humans is needed to confirm the present results, and to determine the potential clinical and public health significance of these findings.

Keywords

Flame retardants; C-reactive protein; blood biomarkers; reactive oxygen species; National Health and Nutrition Examination Survey

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1. Introduction

Polybrominated diphenyl ethers (PBDEs) are found commonly in a number of household and industrial products, in materials like foam padding, textiles, and electronics (ATSDR, 2004). Their application is primarily for flame-retardant purposes. Production of penta- and octa- BDE congeners was discontinued in the US in 2004 (EPA, 2016), but older materials containing these persistent compounds continue to be a source of exposure for humans. PBDEs are not chemically bound to the products in which they are used and hence are readily released into the surrounding environment. Thus, ingestion and inhalation of contaminated dusts are important exposure pathways in humans. The fact that PBDEs are persistent pollutants, accumulating both in the environment and the human body have made them an identified human health concern of high priority by the US Environmental Protection Agency (EPA, 2014).

PBDEs are suspected toxins of the renal, neural, and endocrine systems (Harley et al., 2010; Linares et al., 2015; Meeker et al., 2009; Turyk et al., 2008). *In* utero and/or childhood PBDE exposure has been associated with poorer attention, fine motor coordination, and cognition in school-age children (Eskenazi et al., 2013; Gascon et al., 2011; Herbstman et al., 2010). PBDE concentrations in house dust have been associated with hormonal disturbances in adult men and pregnant women as well (Chevrier et al., 2010; Meeker et al., 2009). Several mechanisms may explain these associations, as illustrated by animal and cellular studies, with thyroid or sex hormone disruption being the primary pathway of interest (Darnerud et al., 2007; Ellis-Hutchings et al., 2006; Hamers et al., 2006; Hamers et al., 2008; Stoker et al., 2005). However, oxidative stress and inflammation may be additional mechanisms by which PBDEs could cause adverse effects in humans. Substantial *in vitro* and animal studies demonstrate that some PBDEs can induce inflammation and oxidative stress (Fernie et al., 2005; He et al., 2008; Park et al., 2014; Peltier et al., 2012). Hydroxylated PBDEs, apparent metabolites of PBDEs, have also been reported to induce oxidative stress in cellular studies (Costa et al., 2014; Usenko et al., 2012; Zhong et al., 2011). Despite this evidence, very few studies to date have examined associations between PBDE exposure and oxidative stress or inflammation in humans. Those that have investigated PBDEs in combination with other persistent organic pollutants, such as polychlorinated biphenyls and p,p-dichlorodiphenyldichloroethylene (DDE) (Kumar et al., 2014), have combined PBDEs and not examined the impact of individual congeners (with the exception of BDE-47) (Turyk et al., 2015), or have addressed this question in study populations without broad generalizability (Rantakokko et al., 2015).

In the present study, we utilized measurements of inflammation and oxidative stress biomarkers in serum or whole blood samples from the National Health and Nutrition Examination Survey (NHANES) from 2003–2004 to explore the relationship between individually measured PBDE congeners and systemic inflammation and oxidative stress. We were limited by examining biomarkers of PBDE exposure that were available in the dataset, which may not include all of the congeners of current interest but does include some that have been examined in other studies, such as 2,2’,4,4’-tetrabromodiphenyl ether (BDE-47). As biomarkers of oxidative stress, we examined bilirubin and gamma-glutamyltransferase (GGT). Decreased levels of bilirubin, a potentially important antioxidant, indicate an

increase in oxidative stress (Stocker et al., 1987); thus, this biomarker has been used in a number of epidemiologic studies (Estrada et al., 2016; Vaishnav et al., 2015; Ferguson et al., 2011). GGT is well-accepted as a biomarker of oxidative stress also (Lee et al., 2004). For inflammation, we examined absolute neutrophil count (ANC), alkaline phosphatase (ALP) and C-reactive protein (CRP). ANC has been used as a sensitive inflammation biomarker in studies examining physiological consequences of environmental exposures (Frost-Pineda et al., 2011; Liao et al., 2005; Yang et al., 2009). Increased ALP (Cheung et al., 2008) and CRP are also commonly used as indicators of overall inflammation and systemic inflammation (Nayeem et al., 2010; Pearson et al., 2003) and may be predictive of adverse health outcomes or indicative of intermediate effects cause by environmental pollutants.

2. Materials and Methods

2.1 Study Participants

NHANES is an ongoing cross-sectional study designed to collect nationally representative data on dietary intake, environmental exposures, and diseases. The present study includes participants and the measurements from the 2003–2004 NHANES cycle. In this analysis, we included 2040 individuals aged 12 and older with measurements available for one or more serum PBDE congener as well as one serum oxidative stress or inflammation biomarker.

2.2 Serum PBDEs

Upon collection of serum from subjects at Mobile Examination Centers (MECs), samples were frozen at −20°C and shipped for analysis to the Division of Environmental Health Laboratory Sciences, National Center for Environmental Health, Centers for Disease Control and Prevention. PBDE congeners measured included: 2,2’, 4-tribromodiphenyl ether (BDE-17); 2,4,4'-tribromodiphenyl ether (BDE-28); 2,3’,4,4’-tetrabromodiphenyl ether (BDE-66); 2,2’,3,4,4’-pentabromodiphenyl ether (BDE-85); 2, 2’,4,4’,5-pentabromodiphenyl ether (BDE-99); 2,2’,4,4’,6-pentabromodiphenyl ether (BDE-100); 2, 2’,4,4’,5,5’-hexabromodiphenyl ether (BDE-153); 2,2’,4,4’,5,6’-hexabromodiphenyl ether (BDE-154); and 2, 2’,3,4,4’,5’,6-heptabromodiphenyl ether (BDE-183).

Measurements of serum PBDEs were made using solid-phase extraction, followed by sample clean up and isotope dilution gas chromatography with high-resolution mass spectrometry, described in detail elsewhere (NCEH, 2011; Sjodin et al., 2008). Quality assurance and quality control (QA/QC) was performed in accordance to the 1988 Clinical Laboratory Improvement Act mandates. Details are recorded in the NHANES Laboratory Procedure Manual of PBDE analytes (NCEH, 2011). The limits of detection (LOD) were variable based on congener and batch, and are examined elsewhere for this dataset (Sjodin et al., 2008). Measurements below the LOD were replaced by the LOD divided by the square root of two (Hornung and Reed, 1990). Because PBDEs are lipid soluble and serum concentrations are dependent upon lipid levels in the blood, we adjusted analyses with serum lipid concentrations to ensure consistently unbiased estimates (Schisterman et al., 2005). Total lipids were estimated by the sum of total cholesterol and triglycerides values, with measurement methods described elsewhere (NCHS, 2008).

Since all PBDEs and outcome biomarker distributions were right-skewed, they were natural-log transformed for analysis. Additionally, PBDEs measurements were lipid standardized by dividing the concentration by serum lipid level for presentation of descriptive statistics and calculation of simple correlations. In regression analyses, unstandardized PBDE levels were used and serum lipid level was included as a covariate in all models (Schisterman et al., 2005).

2.2 Oxidative stress and inflammation biomarkers

At Collaborative Laboratory Services (Ottumwa, IA), GGT, bilirubin, and ALP levels were measured in thawed serum samples in methods described in detail on the NHANES website (NCHS, 2008). Serum CRP was analyzed at the University of Washington using latex-enhanced nephelometry (NCHS, 2000). In MECs at the time of sample collection, ANC was measured in whole blood using the Beckman Coulter MAXM (NCHS, 2007). QA/QC processes were carried out in accordance to the 1988 Clinical Laboratory Improvement Act mandates. Details are recorded in the NHANES Laboratory/Medical Technologists Procedures Manual (available from: https://www.cdc.gov/nchs/data/nhanes/nhanes_03_04/lab.pdf). As with PBDEs, outcome biomarkers were right-skewed and thus ln-transformed for all analyses.

2.3 Covariates

Demographic data were collected from the participants in a survey component of NHANES. These data allowed us to examine age, gender, race/ethnicity, poverty income ratio (PIR, an indicator of socioeconomic status), and education level as potential confounders. Serum cotinine, an indicator of tobacco smoke exposure measured in the laboratory component, and body mass index (BMI), which was part of the physical examination, were examined as covariates as well.

2.4 Statistical analysis

Data analysis was performed using SAS version 9.3 (SAS Institute, Cary, NC). NHANES uses a complex and multistage design in which certain subpopulations are oversampled so that effect estimates within minority groups can be precise. Adjusting regression models for strata, primary sampling units (PSU), and survey weights ensures generalizability to the US population. However, doing so can be inefficient due to over adjustment for variables such as gender, age, race/ethnicity and PIR, which are not only used for weighting but also included as covariates in regression analyses (Korn and Graubard, 1991). Therefore, we constructed both weighted and unweighted models for comparison and presented unweighted results when effect estimates were similar.

In descriptive analyses, we examined distributions of exposure and outcome measurements using geometric means and selected percentiles in the total population. *t* tests and one-way ANOVA were utilized to test differences between groups. Pearson correlation coefficients were calculated to assess relationships among ln-transformed PBDE concentrations, outcome markers, and between the exposures and outcomes. Crude regression models included one PBDE concentration and one oxidative stress or inflammation biomarker. Full multivariable linear regression models included one ln-transformed biomarker as the

dependent variable and one ln-transformed PBDE as a predictor along with age (continuous), gender (dichotomous), race/ethnicity (categorical), BMI (continuous), PIR (continuous), ln-transformed cotinine (continuous) and ln-transformed serum lipid concentration (continuous). Covariates were selected based on associations with exposure and outcome variables in crude analysis and whether they changed effect estimates by greater than 10% upon inclusion in models. All models included the same covariates for consistency and comparability. Regression results were back-transformed to represent a percent change in biomarker in association with an interquartile range (IQR) increase in PBDE concentration.

In secondary analyses, we stratified our data by gender and age group (12–20, 21–40, 41–60, and > 60 years of age) to explore any potential effect modification, as these groups may have different oxidative stress or inflammatory responses to various stimuli. We also examined associations across PBDE quintiles to assess nonlinearity in relationships.

3. Results

Weighted population distributions by categorical covariates are presented in Table 1. Geometric means and selected percentiles of lipid-standardized PBDEs are displayed in Table 2. Because of a high proportion of samples below the LOD (>50%), BDE-17, BDE-85, BDE-66 and BDE-183 were not presented or considered in subsequent analyses. The number of subjects with PBDE data varied from 1,987 up to 2,039 due to missing values for each congener. The geometric means of PBDEs across covariate categories were also examined and consistent with previous studies (results not shown) (Fraser et al., 2009). Geometric means and selected percentiles of oxidative stress and inflammation biomarkers in the study population are displayed in Table 3; each was detected in all samples measured.

Pearson correlation coefficients showed that lipid-standardized PBDE concentrations were moderately to strongly correlated with each other (r =0.56–0.91, p <0.01), as has previously been reported (Fraser et al., 2009). Correlations between oxidative stress and inflammation biomarkers are shown in Supplemental Material Table S1. The oxidative stress biomarkers bilirubin and GGT were weakly but positively correlated (r =0.11, p <0.05). The inflammation marker CRP was inversely correlated with ALP (r =−0.12, p <0.05) but positively correlated with ANC (r =0.24, p <0.05). ALP and ANC were not significantly correlated. Between oxidative stress markers and inflammation markers, GGT had a significant positive association with CRP and bilirubin had a negative correlation with CRP and ANC. The latter association is expected, as bilirubin is thought to represent antioxidant capacity and lower levels indicate reduced capacity, i.e. increased levels of oxidative stress.

Effect estimates from models with and without adjustment for the sampling design were similar; thus, unadjusted results for linear regression models are presented in Table 4. Associations between PBDEs and oxidative stress markers were largely null, although there was a suggestive inverse association between BDE-153 and GGT. In regard to inflammation biomarkers, ALP was significantly and positively associated with BDE-153, and suggestively associated with BDE-99 and BDE-100. ANC was also positively associated

BDE-153 in the adjusted model. CRP was not associated with any of the congeners in adjusted models.

To investigate the possibility of non-linear relationships, we regressed PBDE quintiles on ln-transformed oxidative stress and inflammation markers with adjustment for the same set of covariates included in continuous models. Associations between quintiles of PBDEs and each outcome biomarker are presented in Figure 1 (a–e). As with linear models, few associations were observed between BDE congeners and oxidative stress biomarkers, although there was a suggestive decreasing trend of serum GGT with increasing quintiles for BDE-153 concentrations (p for trend= 0.06). BDE-153 quintiles were associated with increases in both ALP and ANC (p for trends 0.02 and 0.06, respectively), and the association with ALP appeared to be non-monotonic. BDE-100 quintiles were also associated with increases in ALP and ANC (p for trends=0.03). Finally, increasing quintiles of BDE-99 were associated with an increase in CRP (p for trend=0.04).

To examine potential effect modification, we stratified by age groups and gender. There were some differences in adjusted effect estimates between strata with regard to direction and magnitude. Individuals aged 12–20 showed significant inverse associations of small magnitude between GGT and PBDE congeners (BDE-47, BDE-99, BDE-100, BDE-153, BDE-154; Supplemental Material Table S2). In terms of gender strata, males had a significant inverse association between CRP and BDE-153, while in females the association was significant and positive between CRP and congeners BDE-28, BDE-99 and BDE-154 (Supplemental Material Table S3). No other differences were noted.

4. Discussion

We explored the association between a panel of individual serum PBDEs and biomarkers of oxidative stress and inflammation in a large representative US population of adolescents and adults. Of the oxidative stress biomarkers, we observed few associations and in some instances associations opposite of the direction we would expect. However, several inflammation biomarkers were positively associated with serum levels of PBDEs in crude and adjusted models. ALP showed a positive association with BDE-99, BDE-100 and BDE-153. In addition, ANC showed a significant positive association with BDE-153. These associations appeared to be dose-dependent as illustrated in analyses by quintiles of exposure.

Previous studies of the relationships between PBDEs and oxidative stress have been mostly conducted *in vitro* and in animals, and results have varied by PBDE congener and outcome biomarker (Costa et al., 2008). The strongest evidence shows an association between BDE-47 and oxidative stress. This effect has been demonstrated in animal models and in cell lines (An et al., 2011; Costa et al., 2015; Fernie et al., 2005), although a study in zebrafish showed no effect of BDE-47 or other PBDEs on oxidative stress endpoints (Usenko et al., 2015). In regard to inflammation, a cellular study of human first trimester extravillous trophoblasts showed that BDE-47 treatment activated reactive oxygen-mediated inflammatory pathways (Park et al., 2014). We were unable to detect meaningful associations between serum PBDEs and either of these biomarkers in the present analysis,

which is consistent with the results from the few other human studies that have addressed this research question. Kumar et al. examined BDE-47 in relation to lipid or protein related oxidative stress markers and similarly found no associations (Kumar et al., 2014). Likewise, Turyk et al. explored the relationships between BDE-47 and a sum of PBDE congeners with GGT and observed null associations (Turyk et al., 2015). The inability to replicate the animal and *in vitro* findings in humans may be due to lower levels of exposure in humans or differences in metabolism across animal models.

Regarding inflammation, previous *in vitro* and animal studies in various cell types have demonstrated that PBDEs are capable of inducing a proinflammatory response and consequently inflammation. BDE-47 and some other congeners have been shown to cause cytokine release in trophoblast and placental cell lines (Park et al., 2014; Peltier et al., 2012). Our study showed positive associations between PBDE congeners and some biomarkers of inflammation, particularly ANC and ALP, which are consistent with these findings. The only other study to examine BDE-153 alone in relation to inflammation markers in humans observed similar associations between exposure and increased liver inflammation, although that study population focused on individuals undergoing bariatric surgery (Rantakokko, 2015). Also similar to our findings, Ratnakokko et al., Turyk et al. (2015), and Kumar et al. (2014) did not detect any associations between BDE-47 and inflammatory biomarkers in their study populations. These results should be replicated in studies utilizing repeated measures of more specific biomarkers of inflammation.

There were some limitations in our study. First, as is always the case with cross-sectional data, we were unable to establish temporality in the associations observed. Second, single serum measures of oxidative stress and inflammation biomarkers are not optimal as they may vary over time due to diurnal variation (Gu et al., 2009; Lazo et al., 2008). Repeated outcome measurements would improve the power to detect effects. Third, alternative biomarkers should be considered, e.g., cytokines and 8-isoprostane, as circulating levels of these markers may more accurately and thus more sensitively detect inflammation and oxidative stress, respectively, in human populations.

Despite its limitations, this study has many strengths. First, it offers a novel exploration of PBDE associations with oxidative stress or inflammation in a human population, utilizing a large nationally representative population. Second, multiple markers of both oxidative stress and inflammation increased our ability to detect effects, as some of these markers may be more sensitive than others depending on mechanistic pathways that are activated in response to PBDE exposure.

In conclusion, we observed small but significant positive associations between BDE-153 and the inflammation markers ALP and ANC in the general population, suggesting that this congener may be associated with increased inflammation. Additional human studies of oxidative stress and inflammation in relation to PBDE exposure are needed to test these associations, but future research investigating health consequences of PBDE exposure should consider examining health outcomes that are mediated by changes in systemic levels of inflammation.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

Acknowledgments

Funding for this project was provided by the National Institute of Environmental Health Sciences, National Institute of Health (R01ES018872, P42ES017198, P01ES022844, P50ES026049, R01ES022955 and P30ES017885) and by the Intramural Research Program of the National Institutes of Health, National Institute of Environmental Health Sciences.

References

Agency for Toxic Substances and Disease Registry (ATSDR). [accessed 04/12/16] Toxicological Profile for Polybrominated Biphenyls. 2004. Available from: http://www.atsdr.cdc.gov/toxprofiles/tp68.pdf

An J, Yin L, Shang Y, Zhong Y, Zhang X, Wu M, et al. The combined effects of BDE47 and BaP on oxidatively generated DNA damage in L02 cells and the possible molecular mechanism. Mutat Res. 2011; 721:192–198. [PubMed: 21316482]

Cheung BM, Ong KL, Cheung RV, Wong LY, Wat N, Tam S, et al. Association between plasma alkaline phosphatase and C-reactive protein in Hong Kong Chinese. Clinical chemistry and laboratory medicine. 2008; 46:523–527. [PubMed: 18605934]

Chevrier J, Harley KG, Bradman A, Gharbi M, Sjödin A, Eskenazi B. Polybrominated Diphenyl Ether (PBDE) Flame Retardants and Thyroid Hormone during Pregnancy. Environmental Health Perspectives. 2010; 118:1444–1449. [PubMed: 20562054]

Costa LG, de Laat R, Tagliaferri S, Pellacani C. A mechanistic view of polybrominated diphenyl ether (PBDE) developmental neurotoxicity. Toxicology letters. 2014; 230:282–294. [PubMed: 24270005]

Costa LG, Giordano G, Tagliaferri S, Caglieri A, Mutti A. Polybrominated diphenyl ether (PBDE) flame retardants: environmental contamination, human body burden and potential adverse health effects. Acta Biomed. 2008; 79:172–183. [PubMed: 19260376]

Costa LG, Pellacani C, Dao K, Kavanagh TJ, Roque PJ. The brominated flame retardant BDE-47 causes oxidative stress and apoptotic cell death in vitro and in vivo in mice. NeuroToxicology. 2015; 48:68–76. [PubMed: 25797475]

Darnerud PO, Aune M, Larsson L, Hallgren S. Plasma PBDE and thyroxine levels in rats exposed to Bromkal or BDE-47. Chemosphere. 2007; 67:S386–S392. [PubMed: 17257644]

Ellis-Hutchings RG, Cherr GN, Hanna LA, Keen CL. Polybrominated diphenyl ether (PBDE)-induced alterations in vitamin A and thyroid hormone concentrations in the rat during lactation and early postnatal development. Toxicology and Applied Pharmacology. 2006; 215:135–145. [PubMed: 16580039]

Environmental Protection Agency (EPA). [accessed 04/12/16] Technical Fact Sheet –Polybrominated Diphenyl Ethers (PBDEs) and Polybrominated Biphenyls (PBBs). 2014. Available from: https://www.epa.gov/sites/production/files/2014-03/documents/ffrrofactsheet_contaminant_perchlorate_january2014_final_0.pdf

EPA. Polybrominated diphenylethers (PBDEs) Significant New Use Rules (SNUR). 2016 [accessed 08/101/6] Available from: https://www.epa.gov/assessing-and-managing-chemicals-under-tsca/polybrominated-diphenylethers-pbdes-significant-new-use.

Eskenazi B, Chevrier J, Rauch SA, Kogut K, Harley KG, Johnson C, et al. In utero and childhood polybrominated diphenyl ether (PBDE) exposures and neurodevelopment in the CHAMACOS study. Environ Health Perspect. 2013; 121:257–262. [PubMed: 23154064]

Estrada V, Monge S, Gómez-Garre M, Sobrino P, Masiá M, Berenguer J, et al. Relationship between plasma bilirubin level and oxidative stress markers in HIV-infected patients on atazanavir-vs. efavirenz-based antiretroviral therapy. HIV medicine. 2016

Ferguson KK, Loch-Caruso R, Meeker JD. Exploration of oxidative stress and inflammatory markers in relation to urinary phthalate metabolites: NHANES 1999–2006. Environmental science & technology. 2011; 46:477–485. [PubMed: 22085025]

Fernie KJ, Shutt JL, Mayne G, Hoffman D, Letcher RJ, Drouillard KG, et al. Exposure to Polybrominated Diphenyl Ethers (PBDEs): Changes in Thyroid, Vitamin A, Glutathione Homeostasis, and Oxidative Stress in American Kestrels (Falco sparverius). Toxicological Sciences. 2005; 88:375–383. [PubMed: 16120752]

Fraser AJ, Webster TF, McClean MD. Diet contributes significantly to the body burden of PBDEs in the general U.S. population. Environ Health Perspect. 2009; 117:1520–1525. [PubMed: 20019900]

Frost-Pineda K, Liang Q, Liu J, Rimmer L, Jin Y, Feng S, et al. Biomarkers of potential harm among adult smokers and nonsmokers in the total exposure study. Nicotine & Tobacco Research. 2011; 13:182–193. [PubMed: 21330277]

Gascon M, Vrijheid M, Martínez D, Forns J, Grimalt JO, Torrent M, et al. Effects of pre and postnatal exposure to low levels of polybromodiphenyl ethers on neurodevelopment and thyroid hormone levels at 4 years of age. Environment International. 2011; 37:605–611. [PubMed: 21237513]

Gu Y, Zeleniuch-Jacquotte A, Linkov F, Koenig KL, Liu M, Velikokhatnaya L, et al. Reproducibility of serum cytokines and growth factors. Cytokine. 2009; 45:44–49. [PubMed: 19058974]

Hamers T, Kamstra JH, Sonneveld E, Murk AJ, Kester MHA, Andersson PL, et al. In Vitro Profiling of the Endocrine-Disrupting Potency of Brominated Flame Retardants. Toxicological Sciences. 2006; 92:157–173. [PubMed: 16601080]

Hamers T, Kamstra JH, Sonneveld E, Murk AJ, Visser TJ, Van Velzen MJM, et al. Biotransformation of brominated flame retardants into potentially endocrine-disrupting metabolites, with special attention to 2,2′,4,4′-tetrabromodiphenyl ether (BDE-47). Molecular Nutrition & Food Research. 2008; 52:284–298. [PubMed: 18161906]

Harley KG, Marks AR, Chevrier J, Bradman A, Sjödin A, Eskenazi B. PBDE Concentrations in Women's Serum and Fecundability. Environmental Health Perspectives. 2010; 118:699–704. [PubMed: 20103495]

He P, He W, Wang A, Xia T, Xu B, Zhang M, et al. PBDE-47-induced oxidative stress, DNA damage and apoptosis in primary cultured rat hippocampal neurons. NeuroToxicology. 2008; 29:124–129. [PubMed: 18054389]

Herbstman JB, Sjodin A, Kurzon M, Lederman SA, Jones RS, Rauh V, et al. Prenatal exposure to PBDEs and neurodevelopment. Environ Health Perspect. 2010; 118:712–719. [PubMed: 20056561]

Hornung RW, Reed LD. Estimation of Average Concentration in the Presence of Nondetectable Values. Applied Occupational and Environmental Hygiene. 1990; 5:46–51.

Korn EL, Graubard BI. Epidemiologic studies utilizing surveys: accounting for the sampling design. Am J Public Health. 1991; 81:1166–1173. [PubMed: 1951829]

Kumar J, Lind PM, Salihovic S, van Bavel B, Ingelsson E, Lind L. Persistent organic pollutants and inflammatory markers in a cross-sectional study of elderly Swedish people: the PIVUS cohort. Environmental health perspectives. 2014; 122:977. [PubMed: 24911359]

Lazo M, Selvin E, Clark JM. Brief communication: Clinical implications of short-term variability in liver function test results. Annals of Internal Medicine. 2008; 148:348–352. [PubMed: 18316753]

Lee D-H, Blomhoff R, Jacobs DR. Review is serum gamma glutamyltransferase a marker of oxidative stress? Free radical research. 2004; 38:535–539. [PubMed: 15346644]

Liao D, Heiss G, Chinchilli VM, Duan Y, Folsom AR, Lin H-M, et al. Association of criteria pollutants with plasma hemostatic/inflammatory markers: a population-based study. Journal of Exposure Science and Environmental Epidemiology. 2005; 15:319–328.

Linares V, Bellés M, Domingo J. Human exposure to PBDE and critical evaluation of health hazards. Archives of Toxicology. 2015; 89:335–356. [PubMed: 25637414]

Meeker JD, Johnson PI, Camann D, Hauser R. Polybrominated diphenyl ether (PBDE) concentrations in house dust are related to hormone levels in men. Sci Total Environ. 2009; 407:3425–3429. [PubMed: 19211133]

National Center for Environmental Health (NCEH). [accessed 08/10/16] Serum concentrations of polybrominated diphenyl ethers (PBDEs) and polybrominated biphenyl (PBB) in the United States

population: 2003–2004. 2011. Available from: https://www.cdc.gov/nchs/data/nhanes/nhanes_05_06/bfr_met_d_polybrominated_diphenyl_ethers-pbde.pdf

National Center for Health Statistics (NCHS). [accessed 04/12/16] NHANES 2003–2004 Lab Methods. 2008. Available from: http://www.cdc.gov/nchs/nhanes/nhanes2003-2004/lab_methods_03_04.htm

NCHS. [accessed 08/10/16] Laboratory procedure manual: C-reactive protein. 2000. Available from: http://www.cdc.gov/nchs/data/nhanes/nhanes_03_04/l11_c_met_c_reactive_protein.pdf

NCHS. [accessed 08/10/16] Laboratory procedure manual: Complete blood count. 2007. Available from: https://www.cdc.gov/nchs/data/nhanes/nhanes_03_04/l25_c_met_complete_blood_count.pdf

Nayeem F, Anderson KE, Nagamani M, Grady JJ, Lu L-JW. Alkaline phosphatase and percentage body fat predict circulating C-reactive protein in premenopausal women. Biomarkers. 2010; 15:663–670. [PubMed: 20874659]

Park H-R, Kamau PW, Loch-Caruso R. Involvement of reactive oxygen species in brominated diphenyl ether-47-induced inflammatory cytokine release from human extravillous trophoblasts in vitro. Toxicology and Applied Pharmacology. 2014; 274:283–292. [PubMed: 24296301]

Pearson TA, Mensah GA, Alexander RW, Anderson JL, Cannon RO, Criqui M, et al. Markers of inflammation and cardiovascular disease application to clinical and public health practice: a statement for healthcare professionals from the centers for disease control and prevention and the American Heart Association. Circulation. 2003; 107:499–511. [PubMed: 12551878]

Peltier MR, Klimova NG, Arita Y, Gurzenda EM, Murthy A, Chawala K, et al. Polybrominated diphenyl ethers enhance the production of proinflammatory cytokines by the placenta. Placenta. 2012; 33:745–749. [PubMed: 22749501]

Rantakokko P, Männistö V, Airaksinen R, Koponen J, Viluksela M, Kiviranta H, et al. Persistent organic pollutants and non-alcoholic fatty liver disease in morbidly obese patients: a cohort study. Environmental Health. 2015; 14:1. [PubMed: 25564290]

Schisterman EF, Whitcomb BW, Buck Louis GM, Louis TA. Lipid Adjustment in the Analysis of Environmental Contaminants and Human Health Risks. Environmental Health Perspectives. 2005; 113:853–857. [PubMed: 16002372]

Sjodin A, Wong LY, Jones RS, Park A, Zhang Y, Hodge C, et al. Serum concentrations of polybrominated diphenyl ethers (PBDEs) and polybrominated biphenyl (PBB) in the United States population: 2003–2004. Environ Sci Technol. 2008; 42:1377–1384. [PubMed: 18351120]

Stocker R, Yamamoto Y, McDonagh AF, Glazer AN, Ames BN. Bilirubin is an antioxidant of possible physiological importance. Science. 1987; 235:1043–1046. [PubMed: 3029864]

Stoker TE, Cooper RL, Lambright CS, Wilson VS, Furr J, Gray LE. In vivo and in vitro anti-androgenic effects of DE-71, a commercial polybrominated diphenyl ether (PBDE) mixture. Toxicology and Applied Pharmacology. 2005; 207:78–88. [PubMed: 16005038]

Turyk M, Fantuzzi G, Persky V, Freels S, Lambertino A, Pini M, et al. Persistent organic pollutants and biomarkers of diabetes risk in a cohort of Great Lakes sport caught fish consumers. Environmental research. 2015; 140:335–344. [PubMed: 25913152]

Turyk ME, Persky VW, Imm P, Knobeloch L, Chatterton R, Anderson HA. Hormone disruption by PBDEs in adult male sport fish consumers. Environ Health Perspect. 2008; 116:1635–1641. [PubMed: 19079713]

Usenko CY, Abel EL, Kudela M, Janise A, Bruce ED. Comparison of PBDE congeners as inducers of oxidative stress in zebrafish. Environmental Toxicology and Chemistry. 2015:n/a–n/a.

Usenko CY, Hopkins DC, Trumble SJ, Bruce ED. Hydroxylated PBDEs induce developmental arrest in zebrafish. Toxicology and applied pharmacology. 2012; 262:43–51. [PubMed: 22546086]

Vaishnav D, Holkar S, Hivre M. Serum Bilirubin as a Marker of Oxidative Stress in Patients with Hypertension. hypertension. 2015; 5:6.

Yang YJ, Hong Y-C, Oh S-Y, Park M-S, Kim H, Leem J-H, et al. Bisphenol A exposure is associated with oxidative stress and inflammation in postmenopausal women. Environmental research. 2009; 109:797–801. [PubMed: 19464675]

Zhong YF, Wang LL, Yin LL, An J, Hou ML, Zheng KW, et al. Cytotoxic effects and oxidative stress response of six PBDE metabolites on human L02 cells. Journal of Environmental Science and Health, Part A. 2011; 46:1320–1327.

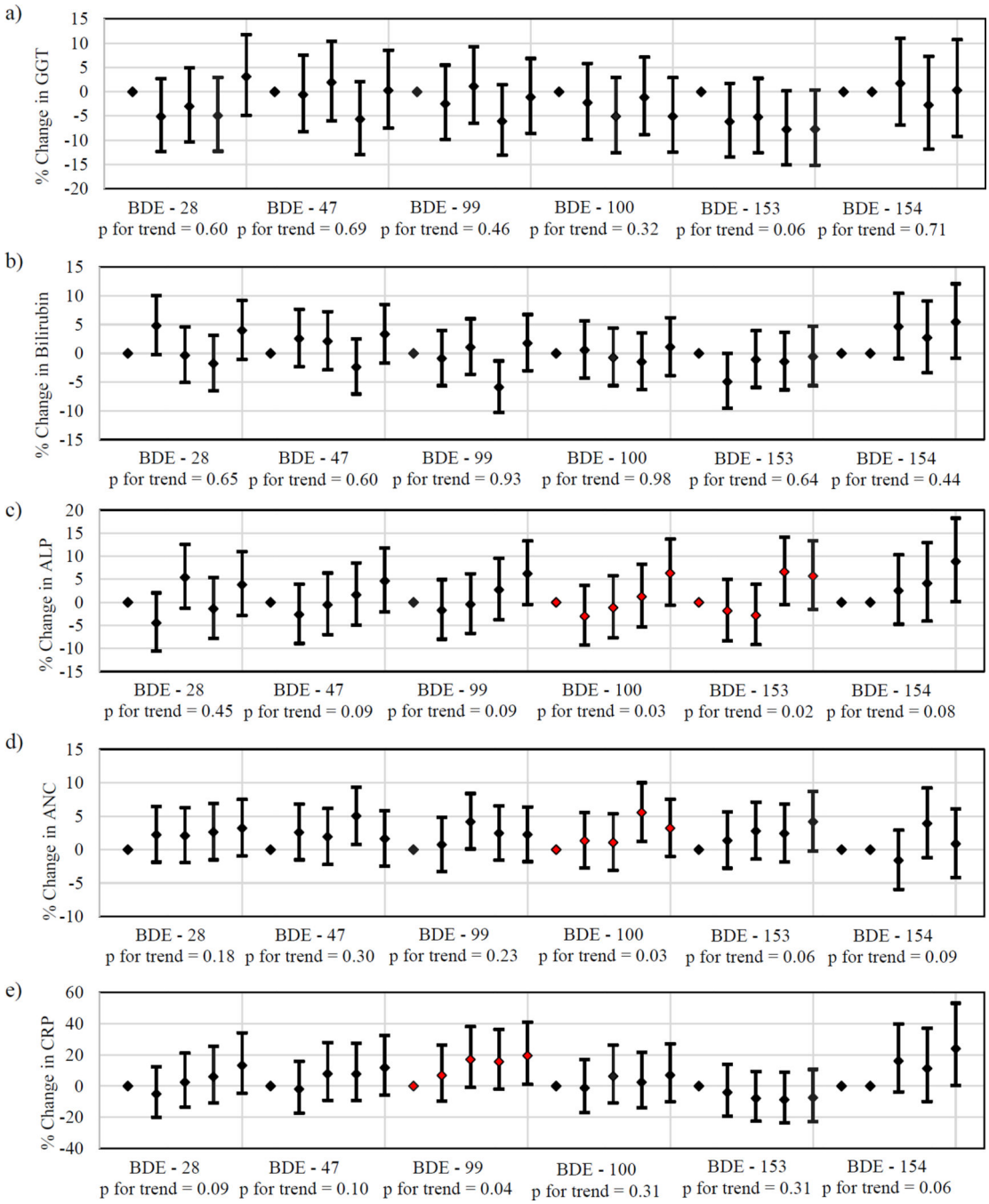

Figure 1.

Adjusteda Percent Change (95% Confidence Intervals) in Oxidative Stress or Inflammmation Biomarker in Association with PBDE quintiles for: a) GGT; b) bilirubin; c) ALP; d) ANC; and e) CRP.

Footnote: Abbreviations: GGT, gamma glutamyl transferase; ALP, alkaline phosphatase; ANC, absolute neutrophil count; CRP, C-reactive protein.

[a]Models adjusted for age, gender, race/ethnicity, poverty income ratio, body mass index, ln-serum cotinine, and ln-serum lipid level.** $p < 0.05$, * $p < 0.10$.

Table 1

Weighted NHANES 2003–2004 Population Characteristics (N = 2040)

Covariate		N (%)
Gender	Male	994 (48.7)
	Female	1046 (51.3)
Age (years)	12 – 20	648 (31.8)
	21 – 40	506 (24.8)
	41 – 60	429 (21.0)
	> 60	457 (22.4)
Race/ethnicity	Mexican American	488 (23.9)
	Other Hispanic	67 (3.28)
	Non-Hispanic White	911 (44.7)
	Non-Hispanic Black	492 (24.1)
	Other Race/multi-racial	82 (4.02)
Body mass index	Underweight (<18.5 kg/m^2)	102 (5.00)
	Normal weight (18.5 – 24.9 kg/m^2)	749 (36.7)
	Overweight (25 – 29.9 kg/m^2)	608 (29.8)
	Obese (>30 kg/m^2)	549 (26.9)
	Missing	32 (1.57)
Poverty income ratio	< 1	472 (23.1)
	1 – 2	502 (24.6)
	2 – 3	295 (14.5)
	> 3	662 (32.5)
	Missing	109 (5.34)
Education (years)	< 12	936 (45.9)
	12	396 (19.4)
	> 12	705 (34.6)
	Missing	3 (0.15)

Table 2

Distribution of PBDE Congener Concentrations Adjusted for Serum Lipid (ng/g lipid).

				Percentiles			
	N	**%>LOD**	**Geometric mean**	**25th**	**50th**	**75th**	**95th**
BDE-28	1987	80.6	1.27	0.55	1.16	2.28	7.87
BDE-47	2016	98.0	23.1	10.8	21.8	45.1	173
BDE-99	2040	70.3	5.60	2.24	4.68	10.4	45.5
BDE-100	1999	94.0	4.36	1.93	4.00	8.27	35.9
BDE-153	2039	93.9	6.08	2.60	5.31	12.1	59.1
BDE-154	2014	55.1	0.66	0.31	0.45	1.00	4.17

Abbreviations: LOD, Limit of Detection (pg/mL)

Table 3

Distribution of Blood Oxidative Stress and Inflammation Markers (N = 2040).

		Percentiles						
	Geometric mean	**5th**	**10th**	**25th**	**50th**	**75th**	**90th**	**95th**
GGT (U/L)	20.1	7.47	8.92	11.8	16.8	26.1	43.6	60.5
Bilirubin (mg/dL)	0.72	0.35	0.41	0.52	0.66	0.84	1.07	1.29
ALP (U/L)	71.1	41.3	46.8	58.2	73.0	95.7	163	255
ANC (1000 cells/uL)	7.05	4.13	4.68	5.58	6.86	8.33	9.99	11.4
CRP (mg/dL)	0.17	0.01	0.02	0.05	0.16	0.42	0.97	1.54

Table 4

Adjusted[a] Percent Change (95% Confidence Interval) in Oxidative Stress or Inflammation Biomarker in Association with an Interquartile Range Increase in PBDE Concentraion

	GGT	Bilirubin	ALP	ANC	CRP
BDE-28	0.57 (−1.41, 2.58)	0.75 (−0.46, 1.98)	0.66 (−0.99, 2.33)	0.26 (−0.76, 1.28)	2.93 (−1.31, 7.35)
BDE-47	−0.19 (−0.85, 0.47)	0.23 (−0.17, 0.64)	0.37 (−0.18, 0.92)	0.05 (−0.29, 0.39)	0.76 (−0.64, 2.18)
BDE-99	−0.15 (−1.43, 1.15)	0.11 (−0.68, 0.91)	0.97 (−0.11, 2.06) *	0.20 (−0.46, 0.87)	2.20 (−0.57, 5.04)
BDE-100	−0.55 (−1.58, 0.49)	0.19 (−0.45, 0.83)	0.84 (−0.03, 1.72) *	0.37 (−0.16, 0.91)	0.58 (−1.62, 2.83)
BDE-153	−0.83 (−1.80, 0.14) *	0.09 (−0.51, 0.70)	0.82 (0.01, 1.65) **	0.53 (0.03, 1.04) **	−1.47 (−3.50, 0.61)
BDE-154	−0.48 (−3.55, 2.68)	0.46 (−1.46, 2.41)	2.03 (−0.59, 4.72)	1.21 (−0.40, 2.85)	5.33 (−1.45, 12.6)

[a]Models adjusted for age, gender, race/ethnicity, poverty income ratio, body mass index, ln-serum cotinine, and ln-serum lipid level.

Abbreviations: GGT, gamma glutamyl transferase; ALP, alkaline phosphatase; ANC, absolute neutrophil count; CRP, C-reactive protein.

** $p < 0.05$,

* $p < 0.10$.